1016 Acute Rejection: Mechanisms, Prediction, and Prevention

Sunday, March 29, 1998, 5:00 p.m.–7:00 p.m.
Georgia World Congress Center, West Exhibit Hall Level
Presentation Hour: 5:00 p.m.–7:00 p.m.

1016-53 Apoptosis and Early Necrosis in Peritransplant Ischemic Injury and Acute Allograft Rejection in Human Heart Transplant Recipients

J.D. Day, R. Gao, U. Narasimhan, S. Gopal, M.T. Day, E.K. Kasper, C.-L. Chen, S.J. Cina, A.L. Robertson, R.H. Kruban. *Stanford University Hospital, Stanford, CA; The Johns Hopkins Medical Institutions, Baltimore, MD, USA*

Background: Cell death may occur by apoptosis or necrosis. Apoptosis is a physiologic process for disposing cells through orderly self destruction, while necrosis results from cytolytic changes following cell injury. We assessed their pattern and extent in multiple samples from 20 patients.

Methods: Apoptosis was detected by in situ end labeling and nuclear chromatin staining and necrosis immunohistochemically using a conjugated myosin light chain monoclonal antibody. Accompanying inflammatory infiltrates were identified by immunoperoxidase staining of lymphocyte subsets. Results were compared with clinical data and routine hematoxylin-eosin evaluation of endomyocardial biopsies.

Results: Apoptosis was identified at the periphery of the ischemic foci in 75% of the biopsies that showed peritransplant ischemic injury by light microscopy, while immunohistochemical staining for myosin light chain (early necrosis) was observed within the cores of all ischemic foci. Apoptotic myocytes were observed in 90% of the biopsies which showed acute rejection and immunohistochemical staining for myosin light chain was observed in 86% of these biopsies. Early myocyte necrosis was detected in 70% of the biopsies evaluated without necrosis by routine histologic grading (Grade 1A - rejection).

Conclusion: Significant myocyte cell death in the form of both apoptosis and early necrosis occurs in peritransplant ischemic injury and in acute allograft rejection. The identification of irreversible intracellular myosin release by the use of monoclonal myosin light chain antibodies in tissue sections proved to be a sensitive technique for detecting the earliest stages of focal myocyte necrosis.

1016-54 The Meaning of Mild Rejection in Steroid-free Heart Transplant Recipients

J.A. Kobashigawa, K. Einhorn, T.K. Ro, J.D. Cassem, J.D. Moriguchi, M.A. Hamilton, A. Hago, N. Kawata, H. Laks. *University of California, Los Angeles, CA, USA*

It has been reported that mild rejection (rej) in heart transplant patients progresses to a more severe form of rej in 10–20% of follow-up biopsies. Many patients are now being weaned from corticosteroids and it has not been established whether mild rej has a similar outcome in these patients. To assess the natural history of untreated mild (ISHLT 1A, 1B) and untreated focal moderate (ISHLT 2) rej in patients off corticosteroids, we reviewed 108 patients transplanted between 4/86 and 9/95 with an average time from transplant to corticosteroid-free immunosuppression of 20 ± 10 months. In these patients, there were 126 episodes of untreated mild and focal moderate rej with outcomes demonstrated in the table.

	Episodes	Next Biopsy		
	N	Cleared	Persisted	Progressed
1A	91	67 (74%)	22 (24%)	2 (2%)
1B	14	1 (7%)	10 (71%)	3 (22%)
2	21	15 (71%)	4 (19%)	2 (10%)

Moderate rej was eventually seen in those "persisted" rej episodes in the following incidence: 1/22 grade 1A episodes, 1/10 grade 1B episodes, and 0/5 grade 2 episodes. Therefore, in patients with 1B rej, a total of 4/13 episodes (31%) progressed to moderate rej which required anti-rejection therapy. During the time of this study, there were 198 abnormal biopsies (including all persisted rejs) out of a total of 845 biopsies performed (23%).

Conclusion: Cardiac transplant patients who are on steroid-free immunosuppression should have careful follow-up if their endomyocardial biopsies demonstrate 1B rejection. ISHLT 1A and 2 biopsy grades appear to have lower risk for progression to moderate rejection. Surveillance biopsies may be necessary to detect rejection in these patients maintained off corticosteroids.

1016-55 Predictive Model to Assess Risk for Coronary Artery Disease and Graft Failure in Cardiac Allograft Recipients: An Immunocytochemical Study

C.A. Labarrere, D.R. Nelson, D.E. Pitts, P.C. Kirlin, H. Halbrook. *Clarian Health (Methodist, Indiana University, Riley Hospitals), Indianapolis, Indiana, USA*

Background: We developed a model for early identification of recipients who subsequently develop transplant-associated coronary artery disease (Tx-CAD) and graft failure.

Methods: Serial biopsies obtained from 121 cardiac allografts (5.5 ± 0.1 biopsies/patient) during the first three months post-transplant were evaluated for deposition of microvascular fibrin, depletion of arteriolar tissue plasminogen activator, presence of arterial/arteriolar endothelial activation markers ICAM-1 and HLA-DR, and changes in vascular antithrombin. An immunocytochemical risk score (IRS) was studied for association with subsequent graft failure and development and progression of Tx-CAD detected using serial coronary angiograms (3.2 ± 0.2/patient).

Results: Allografts with low IRS (score = 0) developed significantly less Tx-CAD ($p < 0.001$) than allografts with moderate (score = 0.5–3.0) or high (score = 3.5–4.0) IRS. The disease progressed in 7%, 18% and 44% of allografts with low, moderate and high IRS, respectively ($p < 0.001$). Allografts with low IRS had significantly less graft failure ($p < 0.001$) than allografts with moderate or high IRS.

Conclusion: Early changes in the microvasculature are associated with impending Tx-CAD and graft failure in cardiac allograft recipients.

1016-56 Detection of Anti-HLA Antibody by Flow Cytometry in Patients With a Left Ventricular Assist Device is Associated With Early Rejection Following Heart Transplantation

D. DeNofrio, F.J. Morales, M. Kamoun, J. Kearns, C. Dorozinsky, B.R. Rosengard, M.A. Acker, E. Loh. *University of Pennsylvania Health System, Philadelphia, PA, USA*

Patients requiring a left ventricular assist device (LVAD) as a bridge to heart transplantation (HT) often have elevated levels of panel reactive antibodies (PRA). The clinical significance of anti-HLA antibodies detected by flow cytometry in PRA negative patients remains unclear. Eighteen patients who underwent LVAD placement as a successful bridge to HT had simultaneous flow cytometry and standard anti-human globulin complement dependent cytotoxicity (AHG-CDC) assays performed to detect anti-HLA antibodies. A positive flow result was defined by a fluorescent ratio of ≥3:1 vs controls. Two patients with a positive PRA and one patient with perioperative death secondary to refractory pulmonary hypertension were excluded. The study population (n = 15) had a mean age of 44 ± 15 yrs, 88% were male, and the mean duration of LVAD support was 94 ± 53 days. In patients with anti-HLA antibodies detected by flow cytometry (n = 6), univariate analysis demonstrated more moderate-severe rejection episodes (ISHLT ≥3A) at 2 months (0.83 ± 0.75 vs 0; $p = 0.04$) and a trend towards decreased time to first rejection (61 ± 17 days vs 225 ± 62 days; $p = 0.06$). No differences in donor recipient HLA mismatch or Kaplan-Meier survival at one year were observed.

Conclusions: Despite a negative PRA, LVAD patients with anti-HLA antibodies detected by flow cytometry have a greater incidence of moderate-severe rejection in the first 2 months following HT. Flow cytometry may be a useful clinical tool in screening PRA negative LVAD patients prior to transplantation. Patients with positive flow cytometry may require more intensive immunosuppression in the early post-HT period.

1016-57 Effective Long-term Methotrexate Therapy for Acute Allograft Rejection After Heart Transplantation

G.M. Mullen, M.A. Silver, C.E. Lawless, J. Mendez, P.C. Barath, K. Malinowska, B.A. Pisani, J.A. Robinson. *Loyola University of Chicago, Heart Transplant Program, Maywood, Illinois, USA*

In spite of progress in immunosuppressive therapy, acute allograft rejection (AAR) still remains a major cause of death and allograft dysfunction after heart transplantation (HT). Standard "triple-drug" immunotherapy (cyclosporine, azathioprine and prednisone) is effective against most, but not all AAR. Other adjunctive agents such as methotrexate (MTX) have been used to decrease frequency and severity of AAR, but its long-term effectiveness is unknown. We retrospectively reviewed 366 HT recipients of whom 34 had received MTX for AAR. We then divided patients into two groups based on AAR grade using a weighed numerical scale, Group I (<3) and Group II (≥3).